

Organic Microchemistry

C. L. OGG

Eastern Regional Research Laboratory, Philadelphia 18, Pa.

DURING the past 2 years there has been a steady flow of papers dealing with quantitative organic microanalytical methods and apparatus. The number of publications in European and Japanese journals has increased, while publications in American journals seem to have declined.

Methods for determining microgram quantities have become numerous and the classification of methods into macro, micro, and ultramicro appears to be crystallizing, with semimicro methods being absorbed mostly in the micro groups.

OXYGEN

Several papers of interest on the determination of oxygen have appeared during the past 2 years. All deal with modifications and sources of error in the Schutze-Unterzaucher carbon reduction method, which is being adopted rapidly by laboratories throughout the country.

One of the chief sources of error in Unterzaucher's modification of Schutze's method has been the interference of hydrogen, especially when samples with low oxygen content are analyzed. To eliminate this source of error Campanile *et al.* (34) included in their unitized apparatus a palladium thimble which permitted the hydrogen to be removed from the combustion products by diffusion. This permitted the sensitive iodometric titration to be retained. Sample weights used ranged from 5 to 50 mg., depending on the oxygen content, and the results were both accurate and precise.

Holowchak and Wear (72) proposed a manometric method which eliminates the error due to hydrogen. The carbon monoxide and hydrogen are oxidized by copper oxide, the water is removed by a dry ice trap, and the carbon dioxide is collected in a liquid nitrogen trap and then measured manometrically. Dundy and Stehr (44) employed a gravimetric determination of the carbon dioxide formed by the iodine pentoxide oxidation as a means of eliminating the large hydrogen error occurring in the analysis of petroleum products with low oxygen content. Gouverneur *et al.* (60) eliminated high blanks and other sources of error by determining the carbon dioxide volumetrically and improving the purity of the transport gas. Hinkel and Raymond (70) used the same approach in their semimicro method. The carbon dioxide was absorbed in 0.05*N* alkali, the carbonate precipitated with barium chloride, and the excess alkali titrated with 0.025*N* acid. Sample weights ranged from 15 to 350 mg. for samples with oxygen contents of 50 to 0.05%. Analysis time was about 45 minutes. Unterzaucher (183) reviewed his micromethod for oxygen, first published in 1940, and gave a comprehensive list of references.

Iron impurities in the carbon are believed by Otting (138) to cause reduction of the quartz tube and give rise to blank values. A new type of high temperature furnace for maintaining the carbon at 1120° C. was described by Steyermark *et al.* (174). The heating element was a 3/8-inch Inconel pipe with insulation around the outside.

A new method for detecting oxygen in organic compounds, based on thermal decomposition and detection of the resulting carbon monoxide or dioxide, was proposed by Goerdeler and Domgörgen (56). A method for determining small quantities of oxygen in nitrogen gas, an important consideration in oxygen analyses, was described by Lourenço (121).

CARBON AND HYDROGEN

In the determination of carbon and hydrogen, the greatest amount of study has been directed toward substitutes for lead dioxide for removing oxides of nitrogen. Kainz (85) changed

from oxygen to air for oxidation of the sample and consequently was able to replace the lead dioxide with a copper section and improve his results. The copper had to be regenerated with hydrogen about every 12 runs.

Lévy and Cousin (115) recommended the use of dry manganese dioxide, previously suggested by Belcher and Ingram. Ingram (77) reported further on the use of manganese dioxide to remove not only oxides of nitrogen but also chlorine and sulfur dioxide, and has measured the conversion of nitrogen to the dioxide in the rapid combustion method.

Lévy and Cousin (115) and Wurzschildt (192) reported the use of silvered pumice in place of silver wire or gauze. Saturated potassium dichromate in concentrated sulfuric acid was used by Mitsui (127), who found this approach satisfactory for samples with NH₂ groups, but NO₂ groups caused the hydrogen values to be high. Klimova and Korshun (100) impregnated dry silica gel with 0.02*N* potassium dichromate in concentrated sulfuric acid until the gel was almost wetted and used this material between two layers of magnesium perchlorate as a dry absorber for oxides of nitrogen.

Berret and Poirier (19) introduced several modifications into the carbon and hydrogen determination, one of which was the elimination of the lead dioxide. Korshun (106) and de Vries and van Dalen (41) reported on the empty-tube method; the former had difficulty in obtaining complete combustion, while the latter authors suggested the use of spark ignition of the vapors to overcome incomplete combustion and explosions. A rapid combustion method using the conventional tube filling was reported by Israelstam (79). Copper instead of lead dioxide was used to eliminate errors due to oxides of nitrogen.

Carbon and hydrogen methods for compounds containing silicon were reported by Klimova *et al.* (101), containing fluorine by Throckmorton and Hutton (181), containing alkali salts by Sirotenko (168), and containing tin and lead by Colaitis and Lesbre (38).

A number of workers have discussed new apparatus and automatic combustion. These include Mitsui (125, 126), who used dry ground joints and no wiping of the absorption tubes, a stationary sample burner, and a hydrogen correction based on the relative humidity and temperature of the room.

Sakamoto (150) discussed automatic apparatus for both the carbon and hydrogen and the Dumas nitrogen determinations. Korshun and Sheveleva (107) found that silver did not remove all the sulfur unless it was heated to near 700° C., that results with and without platinum catalyst were equally good, and that sulfur was retained by the rubber tubing used to join the combustion and absorption tubes. This latter effect was eliminated by the use of ground joints. Teflon joints were used by Kirsten (97) to join the combustion and absorption tubes. Among the several modifications of the carbon and hydrogen apparatus proposed by Sugiyama and Furuhashi (176) was the use of ground joints and the elimination of wiping of the absorption tubes. Hozumi *et al.* (73) described a modified apparatus and the use of a constant air current for combustion. Niederl and Whitman's method of combustion in an atmosphere of nitrogen was tested by Mangeney (123) with good results.

Two wet-combustion procedures for carbon were reported by Peters and Gutmann (144) and Thorn and Shu (180). The former authors' method was for determining radioactive carbon in material with a low carbon content and the latter was a titrimetric procedure.

Kawano *et al.* (90) described the preparation of reagents for Pregl's method, Thompson (179) described a calculator for select-

ing correct sample weights based on the expected analytical values, and Kasagi (89) discussed the application of sequential tests to reduce the number of analyses required for establishing the empirical formulas of organic compounds.

NITROGEN

Dumas. Recent trends in the Dumas method for nitrogen were the modification of the apparatus to permit more rapid burning of the sample and sweeping of the nitrogen into the nitrometer and the use of a mixture of carbon dioxide and oxygen to obtain better sample combustion. Shelberg (164) has made a number of modifications in the Dumas method which obviate slow sample burning and slow gas flow rates. The two-combustion-tube technique has a number of advantages and apparently gives reliable results with an analysis time between 10 and 20 minutes. Gysel (64) combined his previously published method with that of Zimmermann and obtained precise results which appeared to be insensitive to the burning rate. The stationary furnace was maintained at 800° to 850° C. and the sample burner at 950° C. Compounds containing sulfur were most difficult to burn. Mitsui and Tanaka (128) also modified Gysel's earlier method by adding a special nitrometer, using a copper boat, increasing the length of the copper oxide section, and placing the shortened copper layer in two sections.

Ingram (78), Alford (1), and Dirscherl *et al.* (42) all recommended the use of oxygen to aid in the combustion of the sample. In Ingram's method the oxygen prepared electrolytically was mixed with carbon dioxide and the mixture swept through the tube at 15 to 20 ml. per minute. The combustion tube contained only copper for removing the oxygen and the analysis time was about 20 minutes. The method proposed by Alford was more conventional, except for the use of pure oxygen for combustion of the sample. This method proved reliable for refractory heterocyclic compounds. An apparatus for mixing the carbon dioxide and oxygen in any desired proportion was proposed by Dirscherl *et al.* and hopcalite was found necessary if the copper section was located in the front of the tube filling. Kirsten (95) claimed that free oxygen in the presence of the copper oxide caused nitrogen oxide retention errors. He recommended the use of nickel oxide because it can be used at higher temperatures and does not liberate oxygen on heating, as does copper oxide. Pagel and Oita (139) discussed the errors in the Dumas method and particularly those due to air absorption.

The preliminary collaborative study of the Dumas method by the Association of Official Agricultural Chemists (135) showed a great variability in procedures throughout the country but little difference in accuracy and precision that could be associated with specific steps or techniques used by the various collaborators. Long furnace temperatures higher than 750° C. produced more precise results for the samples containing N—N and N—O bonds. Results obtained by the newer procedures proposed by Shelberg, Zimmermann, and Kirsten compared very favorably with those by the more conventional procedures, but too few collaborators used these new methods to permit a very reliable comparison.

Four articles have been published on carbon dioxide generators, one by Childs and Moore (36) on a modified Poth-type generator, and one by Konovalov (104) on preparation of the Kipp generator. Parkin *et al.* (140) and the British Standards Institute (26) discussed the use of solid carbon dioxide as the gas source.

Kjeldahl. The most prominent notes with regard to the Kjeldahl method for nitrogen have been the effect of the temperature of the digestion mixture on the digestion time and recovery of nitrogen and the design of new distillation apparatus. In the collaborative study reported by Lake (110) it was shown that the temperature could be raised by adding potassium sulfate to 410° C. without causing loss of nitrogen. The method that proved satisfactory for petroleum oils was 15 to 500 mg. of sample, 3 grams of potassium sulfate, 0.1 gram of mercuric oxide, and 4 ml. of concentrated sulfuric acid plus 0.5 ml. of acid for each 100

mg. above 100 mg. of sample with digestion for 1 to 1.5 hours at a temperature not greater than 380° C. The results indicated that the temperature should not be far below this value. Ribas and Vázquez-Gesto (147) found that refractory heterocyclic compounds could be digested in 30 minutes if 3 grams of potassium sulfate in 2 ml. of sulfuric acid with mercury and selenium catalysts were used. The sealed-tube method of White and Long was used for ultramicroanalysis by Grunbaum *et al.* (62). A digestion temperature of 450° C. produced good recoveries, whereas temperatures of 470° C. and higher caused some decomposition of the ammonia.

Fish (50), Konovalov (105), Pepkowitz (142), and Ogg and Willits (137) all reported on the analyses of materials containing N—N and N—O linkages. Fish used reduction with zinc and hydrochloric acid in acetic acid or methanol, while Konovalov heated the materials in sealed tubes with hydriodic acid. The iodine liberated was fixed with red phosphorus and removed by boiling with sulfuric acid. Pepkowitz found that azide nitrogen could be determined if sodium thiosulfate was added to the sample before the sulfuric acid. The collaborative work reported by Ogg and Willits showed that about half the analysts obtained good results for compounds with N—N and N—O linkages by refluxing with hydriodic acid prior to digestion, whereas the results of the other analysts were low. As all analysts were following the same procedure, it must be concluded that some step or direction given in the method was not sufficiently well defined. Marzadro (124) claimed to be able to distinguish between various forms of nitrogen in heterocyclic compounds on the basis of results obtained by the Dumas-Zimmermann method and by a modified Kjeldahl procedure.

Titrimetric methods for determining nitrogen without distillation were proposed by Harvey (66), Lestra and Roux (112), and Llacer (117). The first two were based on the titration of liberated iodine with sodium thiosulfate, and the last method used the technique of Marcali and Rieman.

Seligson and Seligson (162) substituted discarded penicillin bottles for Conway cells and determined the nitrogen with Nessler's reagent. Slavík and Smetana (169) used Nessler's reagent to determine the ammonia liberated by enzymic reactions, the ammonia being absorbed in a tuft of glass wool soaked in 4N sulfuric acid.

The British Standards Institute (29) has published specifications for the micro-Kjeldahl digestion and distillation apparatus. Distillation apparatus, both micro and semimicro, was the subject of papers by Kirsten (91), Jenden and Taylor (80), Sickels and Schultz (166), Sheers and Cole (163), and Nógrády (131).

An interesting modification of the Van Slyke method for the microdetermination of amino nitrogen was proposed by Hussey and Maurer (75). The gases evolved were swept through a chromic acid-alkaline permanganate absorption train to remove oxides of nitrogen and the nitrogen gas was caught and measured in a micronitrometer.

SULFUR

Collaborative studies (134, 136) have been made of the more common micromethods for determining sulfur, including Carius, Parr, and catalytic combustions and both volumetric and gravimetric determination of the sulfate formed. The Carius and catalytic methods were about equally good, were more widely used, and appeared to be better than the Parr bomb procedure. More precise results were obtained by the volumetric than by the gravimetric method tested. Specifications for the micro Grote combustion train for sulfur and the halogens have been published by the British Standards Institute (28). Zimmermann (194) defended and elaborated on his method for sulfur, which involves reduction by potassium, distillation of the hydrogen sulfide, oxidation with iodate, and titration of the excess iodate. Kirste and Carstens (98) claimed that errors occurred during the decomposition of the sample in Zimmermann's method and that these

were approximately compensated by errors in the titration. Another titrimetric method for determining hydrogen sulfide or sulfide sulfur in microgram quantities was proposed by Dunicz and Rosenqvist (45). The method was based on the oxidation by alkali hypochlorite in highly alkaline solution.

Belcher *et al.* (15) determined sulfate alkalimetrically after precipitation with 4-amino-4'-chlorodiphenyl to eliminate errors due to nitrates or halides. In the titrimetric determination of sulfate with rhodizonate indicator Scalandre and Guerrero (154) suggested adjusting the solution to less than pH 4 to eliminate phosphate interference.

Szekeres *et al.* (178) proposed a wet-digestion procedure using dichromate and nitric acid to convert sulfur to the sulfate, followed by gravimetric determination as the barium salt. Oda *et al.* (133) combined Stragand's method for sulfur with the usual carbon and hydrogen determination to produce a procedure for simultaneous determination of the three elements. Fischer (49), using an electron microscope, studied the effect of precipitating conditions on the nature of the barium sulfate crystals.

A spectrophotometric method for determining microgram amounts of sulfur in plant materials, etc., was described by Johnson and Nishita (81). The method involves the reduction of sulfates to hydrogen sulfide with hydriodic acid, formic acid, and red phosphorus, and the conversion of the hydrogen sulfide to methylene blue.

Toennies and Bakay (182) and Grassner (61) reported turbidimetric methods for determining microgram quantities of sulfur. The former authors used a glycol-ethyl alcohol-water system to stabilize the barium sulfate suspension and were able to determine accurately as little as 3 γ of sulfur.

A rapid combustion process for sulfur and halogens was described by Belcher and Ingram (14) and an improved quartz tube combustion apparatus by Peters *et al.* (143). The effect of varying conditions on the combustion of sulfanilamide in the Grote apparatus was described by Sundberg and Royer (177).

HALOGENS

In the collaborative studies on the determination of chlorine and bromine conducted by Steyermark and Faulkner (172) and Steyermark and Garner (173), better results were obtained by the Carius method of combustion followed by gravimetric determination of the halide. The preliminary study (172) showed that most of the collaborators were using either Carius or catalytic combustion methods and that these were preferable to the Parr bomb method. In the second study the results favored the Carius over the catalytic method tested, in both accuracy and precision. Specifications for a halogen and sulfur combustion train (Pregl) have been published by the British Standards Institute (27).

Although considerable work is still being done to devise titrimetric procedures for the halogens, no single method has gained sufficient favor to make it outstanding among the many methods proposed. Mercurimetric titration seems to be gaining in favor, and Jureček and Večeřa (83) have reported that with diphenylcarbazone as indicator, acidity under normal conditions has no deleterious effect on the results.

A new approach to argentometric titration reported by Hasselmann and Laustriat (68) recommended illuminating the solution with a narrow light beam during titration. The color of the pencil of light changed sharply from colorless or yellow to red at the equivalence point. For argentometric titrations, Dean *et al.* (40) recommended the use of polyethylene glycol 400 as a protective colloid. Rao and Shah (146) modified the Zacherl-Krainick wet-digestion method to permit the alkalimetric determination of the halogens in compounds containing nitrogen and sulfur. Another volumetric procedure proposed by Flaschka (51) required an ethylenediaminetetraacetic acid titration of nickel liberated from ammoniacal nickel tetracyanide by the silver salts. Nutten (132) found the mercuric oxycyanide method preferable to

the direct alkalimetric method for halogens. Belcher and Goulden (13) tested silver, mercurous, and mercuric iodates for the determination of the chloride ion and found the three to be relatively comparable, with errors up to 2%. The iodate liberated by the chloride ion was titrated iodometrically.

Decomposition of the halogen-containing material with an alkali metal was investigated by Lohr *et al.* (118) and Kainz and Resch (86). The former authors used sodium metal in a nickel bomb and found the gravimetric analyses more accurate than the volumetric method; the latter used potassium in a glass tube and followed this with a Volhard titration.

Kainz (84) also proposed a procedure for bromine involving disintegration with potassium, followed by oxidation and iodometric titration. Simultaneous determination of chlorine and bromine was accomplished by Lévy (114) by combining argentometry and potentiometry. Bromine values had to be corrected by a value related to the quantity of chlorine present. A rapid potentiometric method for low concentrations of chloride ion employing a double silver-silver chloride electrode system was proposed by Blaedel *et al.* (21). Kirsten (96) made the oxygen and combustion products from a dry catalytic combustion react with hydrogen to effect more complete absorption, and give a higher concentration of halide with less foreign ions. A potentiometric determination was recommended. Cheek (35) and Pirt and Chain (145) applied the Conway microdiffusion technique to the determination of halogens, the latter authors using fuming nitric acid with silver nitrate to liberate the bound halogen. Gordon (58), after liberating the halogen with permanganate oxidation, applied a more sensitive colorimetric modification of the Conway microdiffusion technique. Another colorimetric method based on precipitation of chloride as mercurous chloride and measurement of the excess mercury was proposed by Staemmler (171). An apparatus for rapid combustion of compounds containing halogens and sulfur was described by Belcher and Ingram (14) along with suggested procedures for completing the determinations.

Zacherl and Stöckl (193) recommended the use of bromine-glacial acetic acid as absorbent in the Leipert method for iodine and also advised heating the solution after reduction of the bromine with formic acid. Lein and Schwartz (111) discussed the use of ceric sulfate-arsenious acid reaction for the microdetermination of iodine, while Bosch and Rubia Pacheco (22) recommended that photometric measurements of iodine be made in carbon disulfide solution rather than in water.

The micromethod for fluorine proposed by Belcher *et al.* (11) involves fusion with alkali metal in a nickel bomb, titration with thorium nitrate, and back-titration of a comparison solution with standard sodium fluoride.

Three papers by Ballczo and Kaufmann (7-9) deal with a method for the determination of microgram quantities of fluorine and its application. They recommended the addition of methylene blue to the alizarin sulfonate indicator to increase the sharpness of the end-point color change.

OTHER ELEMENTS

Three methods for determining microgram quantities of phosphorus have been described by Harvey (67), Nakamura (129), and Schaffer *et al.* (155). All three are spectrophotometric procedures involving phosphomolybdic acid. In Schaffer's method the phosphomolybdic acid is extracted from the reaction mixture with octyl alcohol and determined by capillary spectrophotometry. A new microprocedure for phosphorus has been proposed by Ma and McKinley (122).

Methods for determining selenium in organic compounds were described by Gould (59), who completed the analysis titrimetrically, and by Kan (88), who used a gravimetric method. Bricker and Sweetser (25) in the determination of arsenic determined the end point of the titration spectrophotometrically at 320 m μ . The use of absorption in the ultraviolet for the detection of photo-

metric end points seems to be an unexplored field and offers the advantages of eliminating indicators and indicator errors as well as the necessity of titrating to an exact end point. A comprehensive review of methods for determining metals in organic compounds was made by Belcher *et al.* (12), and both gravimetric and colorimetric micromethods for alkali metals were described by Duval (46). Van Etten and Wiele (187) proposed the use of ion exchange resins for determining organic salts.

GROUP ANALYSIS

Sodium thiosulfate was shown by Franzen *et al.* (52) to react with ethyl iodide under the conditions of the alkoxyl or alkylimide analysis, whereas the extent of the reaction with methyl iodide was considerably less. The difference in the reactivity of the two iodides was suggested as a means of distinguishing between the two. According to Huang and Morsingh (74), fully methylated dibenzyls have an anomalous behavior in the Zeisel method, in that they liberate methyl iodide although no methoxyl is present.

Kirsten and Stenhagen (99) studied the Kuhn-Roth method of C-methyl groups and proposed certain changes to improve the method. Three new apparatus for determining unsaturation by catalytic hydrogenation were described by Savacool and Ullyot (153), Vandenheuvel (185), and Schöniger (156). Micromethods for carbonyl groups were described by Bennett *et al.* (17), Schöniger and Lieb (159), and Schöniger *et al.* (160).

Determination of carboxy groups in aromatic compounds by decarboxylation was discussed by Beroza (18). Smith *et al.* (170) described the determination of acids, bases, and esters in small sealed tubes or serum bottles. The technique is versatile and promises to have many applications. Microgram quantities of fatty acids were titrated potentiometrically by Grunbaum *et al.* (63) and the fatty acids in serum were determined by Kaiser and Kagan (87).

A spectrophotometric micromethod for polyunsaturated fatty acids was described by Herb and Riemenschneider (69). Van Etten (186) described a reliable yet simple procedure for the microdetermination of saponification equivalents. Methods for the microdetermination of sulfhydryl groups in proteins were proposed by Kolb and Toennies (103) and by Sahashi and Shibasaki (149). Determination of aromatic hydroxyl groups with dibromoaminophenol was described by Shishikura (165). Brunner and Thomas (33) found that phenolic acetates hydrolyze rather rapidly during hydrolysis of the anhydride in the acetylation method of hydroxyl analysis. They recommended the use of 20- to 80-mg. samples and only a 3-minute hydrolysis.

APPARATUS

No attempt has been made to include references to all microapparatus described during the past 2 years. In general, only those pieces having a fairly direct relationship to the methods already discussed are included.

Two automatic combustion apparatus (116, 126) as well as a high temperature furnace for organic elemental analysis (93) have been described. The British Standards Institute published specifications for platinum, nickel, and porcelain crucibles (30) and for combustion boats, sheath, and contact stars (31). Accessory apparatus for the combustion of hygroscopic liquids (57) and for aqueous solutions of organic compounds (82) were described. Stainless steel absorption tubes (109) were used in place of glass tubes in the semimicrodetermination of carbon and hydrogen. Two apparatus for determining the molecular weight of volatile materials (39, 130) and a modified Signer apparatus (188) were described.

Two articles on microburets include descriptions of a high-precision buret (54) and the construction and calibration of simple semiautomatic microburets (184). Micropipets (6, 10, 20, 37, 157, 161) of various design and operation have been described.

Balances. Quartz fiber balances, their construction, maxi-

mum load, sensitivity, reproducibility, and application have been the subject of four papers (4, 24, 108, 152). Preference of the analyst for certain numbers has been shown by a statistical study (65) to be the cause of errors in weighing with the microbalance. Tests with an Ainsworth and a Kuhlman balance (120) showed that the drift of the zero reading was excessive under the conditions tested. An Ainsworth assay balance (32) was found to be less sensitive to temperature fluctuations than microbalances but more affected by vibrations. Microanalysis using a Mettler microanalytical balance has been discussed (71) and the present position of microbalances reviewed (167).

GENERAL AND REVIEWS

A discussion of the accuracy of organic microanalysis (94) indicates that the error is more often due to the method than to the analyst. General discussions have been published on the application of amperometric titrations (141), on errors in amperometric and starch-indicator methods (23), on ultramicromethods, apparatus, and techniques (2, 47, 148), and on recent developments in microanalysis (92, 102) and microapparatus (76, 175).

Reference materials or standards for all elements and groups commonly determined have been listed (5) and some new commercial products of interest to microchemists have been discussed (16). The work of two committees, one on the standardization of apparatus, the other on methods, was reviewed (189). Micromethods applicable to medicinal substances were described (119) and microanalysis and chemistry of specific, selective, and sensitive reactions were discussed (48).

A number of review articles concerning or related to organic microanalysis have been published during the past 2 years. Among them are an appraisal of microchemistry (190), a discussion of the progress and possibilities of organic microanalysis (113), a review of some new methods (43), a historical review of microchemistry (53), two general reviews on microchemistry (3, 158), and one on semimicrotechniques (191). Microanalytical methods for proteins in blood plasma (151) and for clinical analysis (55) have also been reviewed.

LITERATURE CITED

- (1) Alford, W. C., *ANAL. CHEM.*, **24**, 881-4 (1952).
- (2) Alimarin, I. P., and Petrikova, M. N., *Zhur. Anal. Khim.*, **7**, 341-8 (1952).
- (3) Alvarez Querol, M. C., *Inform. quim. anal. (Madrid)*, **5**, 209-17 (1951).
- (4) Ambrosino, C., *Chimica e industria (Milan)*, **33**, 775-7 (1951).
- (5) *Analyst*, **78**, 258-9 (1953).
- (6) Anderson, H. H., *ANAL. CHEM.*, **24**, 579-83 (1952).
- (7) Balczon, H., and Kaufmann, O., *Mikrochimie ver. Mikrochim. Acta*, **38**, 237-57 (1951).
- (8) *Ibid.*, **39**, 9-12 (1952).
- (9) *Ibid.*, pp. 13-21.
- (10) Barth, L. G., and Sze, L. C., *Rev. Sci. Instr.*, **22**, 978-80 (1951).
- (11) Belcher, R., Caldas, E. F., Clark, S. J., and Macdonald, A., *Mikrochim. Acta*, **1953**, 283-9.
- (12) Belcher, R., Gibbons, D., and Sykes, A., *Mikrochimie ver. Mikrochim. Acta*, **40**, 76-103 (1952).
- (13) Belcher, R., and Goulden, R., *Mikrochim. Acta*, **1953**, 290-7.
- (14) Belcher, R., and Ingram, G., *Anal. Chim. Acta*, **7**, 319-23 (1952).
- (15) Belcher, R., Nutten, A. J., and Stephen, W. I., *Mikrochim. Acta*, **1953**, 51-7.
- (16) Benedetti-Pichler, A. A., *Mikrochimie ver. Mikrochim. Acta*, **39**, 319-21 (1952).
- (17) Bennett, A., May, L. G., and Gregory, R., *J. Lab. Clin. Med.*, **37**, 643 (1951).
- (18) Beroza, M., *ANAL. CHEM.*, **25**, 177-9 (1953).
- (19) Berret, R., and Poirier, P., *Bull. soc. chim. France*, **1951**, 724-7.
- (20) Birket-Smith, E., *Scand. J. Clin. Lab. Invest.*, **3**, 234-5 (1951).
- (21) Blaedel, W. J., Lewis, W. B., and Thomas, J. W., *ANAL. CHEM.*, **24**, 509-12 (1952).
- (22) Bosch, F. de A., and Rubia Pacheco, J. de la, *Anales real soc. españ. fis. y quím.*, **47B**, 263-8 (1951).
- (23) Bradbury, J. H., and Hambly, A. N., *Australian J. S. Research*, **A5**, 541-54 (1952).
- (24) Bradley, R. S., *J. Sci. Instr.*, **30**, 84-9 (1953).

- (25) Bricker, C. E., and Sweetser, P. B., *ANAL. CHEM.*, **24**, 409-11 (1952).
- (26) Brit. Standard 1428, Pt. A2 (1952).
- (27) *Ibid.*, Pt. A3 (1952).
- (28) *Ibid.*, Pt. A4 (1953).
- (29) *Ibid.*, Pt. B1 (1953).
- (30) *Ibid.*, Pt. E1 (1953).
- (31) *Ibid.*, Pt. I1 (1953).
- (32) Bromund, W. H., and Benedetti-Pichler, A. A., *Mikrochemie ver. Mikrochim. Acta*, **38**, 505-13 (1951).
- (33) Brunner, H., and Thomas, H. R., *J. Appl. Chem. (London)*, **3**, 49-54 (1953).
- (34) Campanile, V. A., Badley, J. H., Peters, E. D., Agazzi, E. J., and Brooks, F. R., *ANAL. CHEM.*, **23**, 1421-6 (1951).
- (35) Cheek, D. B., *J. Appl. Physiol.*, **5**, 639-45 (1953).
- (36) Childs, C. E., and Moore, V. A., *ANAL. CHEM.*, **25**, 204 (1953).
- (37) Claff, C. L., *Science*, **105**, 103-4 (1947).
- (38) Colaitis, D., and Lesbre, M., *Bull. soc. chim. France*, **1952**, 1069-72.
- (39) Colson, A. F., *Analyst*, **77**, 139-43 (1952).
- (40) Dean, R. B., Wiser, W. C., Martin, G. E., and Barnum, D. W., *ANAL. CHEM.*, **24**, 1638-9 (1952).
- (41) de Vries, G., and van Dalen, E., *Anal. Chim. Acta*, **7**, 274-6 (1952).
- (42) Dirscherl, A., Padowetz, W., and Wagner, H., *Mikrochemie ver. Mikrochim. Acta*, **38**, 271-2 (1951).
- (43) Dory, I., and Messmer, A., *Magyar Kém. Lapja*, **6**, 66-71 (1951).
- (44) Dundy, M., and Stehr, E., *ANAL. CHEM.*, **23**, 1408-13 (1951).
- (45) Dunicz, B. L., and Rosenqvist, T., *Ibid.*, **24**, 404-6 (1952).
- (46) Duval, C., *Chim. anal.*, **34**, 209-21 (1952).
- (47) El-Badry, H. M., and Wilson, C. L., *Mikrochemie ver. Mikrochim. Acta*, **40**, 141-59 (1952).
- (48) Feigl, F., *Ibid.*, **39**, 368-88 (1952).
- (49) Fischer, R. B., *ANAL. CHEM.*, **23**, 1667-71 (1951).
- (50) Fish, V. B., *Ibid.*, **24**, 760-2 (1952).
- (51) Flaschka, H., *Mikrochemie ver. Mikrochim. Acta*, **40**, 21-6 (1952).
- (52) Franzen, F., Disse, W., and Eysell, K., *Mikrochim. Acta*, **1953**, 44-50.
- (53) Gillis, J., *Mededeel. Vlaam. Chem. Ver.*, **12**, 15-22 (1950).
- (54) Gilmont, R., *ANAL. CHEM.*, **25**, 1135 (1953).
- (55) Glick, D., *Chem. Eng. News*, **31**, 139-42 (1953).
- (56) Goerdeler, J., and Domgörgen, H., *Mikrochemie ver. Mikrochim. Acta*, **40**, 212-17 (1953).
- (57) Goldberger, H., and Pöhm, M., *Ibid.*, **39**, 73-4 (1952).
- (58) Gordon, H. T., *ANAL. CHEM.*, **24**, 857-62 (1952).
- (59) Gould, E. S., *Ibid.*, **23**, 1502-3 (1951).
- (60) Gouverneur, P., Schreuders, M. A., and Degens, P. N., Jr., *Anal. Chim. Acta*, **5**, 293-312 (1951).
- (61) Grassner, F., *Z. anal. Chem.*, **135**, 186-91 (1952).
- (62) Grunbaum, B. W., Schaffer, F. L., and Kirk, P. L., *ANAL. CHEM.*, **24**, 1487-90 (1952).
- (63) *Ibid.*, **25**, 480-2 (1953).
- (64) Gysel, H., *Helv. Chim. Acta*, **35**, 802-7 (1952).
- (65) Gysel, H., *Mikrochim. Acta*, **1953**, 266-82.
- (66) Harvey, H. W., *Analyst*, **76**, 657-60 (1951).
- (67) *Ibid.*, **78**, 110-14 (1953).
- (68) Hasselmann, M., and Laustriat, G., *Compt. rend.*, **234**, 625-7 (1952).
- (69) Herb, S. F., and Riemenschneider, R. W., *ANAL. CHEM.*, **25**, 953-5 (1953).
- (70) Hinkel, R. D., and Raymond, R., *Ibid.*, **25**, 470-9 (1953).
- (71) Hirayama, H., Ieki, T., and Miyahara, K., *J. Pharm. Soc. Japan*, **73**, 103-4 (1953).
- (72) Holowchak, J., and Wear, G. E. C., *ANAL. CHEM.*, **23**, 1404-7 (1951).
- (73) Hozumi, K., Imaeda, K., and Tanaka, M., *J. Pharm. Soc. Japan*, **72**, 658-61 (1952).
- (74) Huang, R. L., and Morsingh, F., *ANAL. CHEM.*, **24**, 1359 (1952).
- (75) Hussey, A. S., and Maurer, J. E., *Ibid.*, **24**, 1642-4 (1952).
- (76) Ingram, G., *Chemist and Druggist*, **155**, 615-17 (1951).
- (77) Ingram, G., *Mikrochim. Acta*, **1953**, 71-8.
- (78) *Ibid.*, pp. 131-47.
- (79) Israelstam, S. S., *ANAL. CHEM.*, **24**, 1207-9 (1952).
- (80) Jenden, D. J., and Taylor, D. B., *Ibid.*, **25**, 685-6 (1953).
- (81) Johnson, C. M., and Nishita, H., *Ibid.*, **24**, 736-42 (1952).
- (82) Jones, A. R., *Rev. Sci. Instr.*, **24**, 230 (1953).
- (83) Jureček, M., and Večeřa, M., *Chem. Listy*, **46**, 620-2 (1952).
- (84) Kainz, G., *Mikrochemie ver. Mikrochim. Acta*, **38**, 124-31 (1951).
- (85) *Ibid.*, **39**, 166-75 (1952).
- (86) Kainz, G., and Resch, A., *Ibid.*, **39**, 1-8 (1952).
- (87) Kaiser, E., and Kagan, B. M., *ANAL. CHEM.*, **23**, 1879-80 (1951).
- (88) Kan, M., *Ann. Repts. Takeda Research Lab.*, **11**, 54-9 (1952).
- (89) Kasagi, M., *Japan Analyst*, **1**, 148-51 (1952).
- (90) Kawano, T., Yamamoto, S., and Nakayama, M., *Ibid.*, **1**, 126-30 (1952).
- (91) Kirsten, W., *ANAL. CHEM.*, **24**, 1078 (1952).
- (92) *Ibid.*, **25**, 74-86 (1953).
- (93) *Ibid.*, pp. 805-6.
- (94) Kirsten, W., *Anal. Chim. Acta*, **5**, 489-93 (1951).
- (95) Kirsten, W., *Mikrochemie ver. Mikrochim. Acta*, **40**, 121-37 (1952).
- (96) *Ibid.*, pp. 170-2.
- (97) Kirsten, W., *Mikrochim. Acta*, **1953**, 41-3.
- (98) Kirsten, W., and Carstens, C., *Anal. Chim. Acta*, **5**, 272-6 (1951).
- (99) Kirsten, W., and Stenhagen, E., *Acta Chem. Scand.*, **6**, 682-9 (1952).
- (100) Klimova, V. A., and Korshun, M. O., *Zhur. Anal. Khim.*, **6**, 230-3 (1951).
- (101) Klimova, V. A., Korshun, M. O., and Bereznitskaya, E. G., *Doklady Akad. Nauk S.S.S.R.*, **84**, 1175-8 (1952).
- (102) Kohn, M., *Anal. Chim. Acta*, **5**, 337-44 (1951).
- (103) Kolb, J. J., and Toennies, G., *ANAL. CHEM.*, **24**, 1164-9 (1952).
- (104) Kononov, A., *Ind. chim. belge*, **16**, 209-16 (1951).
- (105) *Ibid.*, **18**, 329-39 (1953).
- (106) Korshun, M. O., *Zhur. Anal. Khim.*, **7**, 96-100 (1952).
- (107) Korshun, M. O., and Sheveleva, N. S., *Ibid.*, **7**, 104-11 (1952).
- (108) Kuck, J. A., Altieri, P. L., and Towne, A. K., *Mikrochim. Acta*, **1953**, 254-65.
- (109) Kuck, J. A., and Arnold, M., *Mikrochemie ver. Mikrochim. Acta*, **38**, 521-31 (1951).
- (110) Lake, G. R., *ANAL. CHEM.*, **24**, 1806-11 (1952).
- (111) Lein, A., and Schwartz, N., *Ibid.*, **23**, 1507-10 (1951).
- (112) Lestra, H., and Roux, G., *Compt. rend.*, **233**, 1453-5 (1951).
- (113) Lévy, R., *Bull. soc. chim. France*, **1952**, 672-87.
- (114) Lévy, R., *Compt. rend.*, **235**, 882-4 (1952).
- (115) Lévy, R., and Cousin, B., *Bull. soc. chim. France*, **1952**, 728-36.
- (116) Lévy, R., and Mathieu, P., *Ibid.*, **1952**, 737-40.
- (117) Llacer, A. J., *Mikrochemie ver. Mikrochim. Acta*, **40**, 173-8 (1952).
- (118) Lohr, L. J., Bonstein, T. E., and Frauenfelder, L. J., *ANAL. CHEM.*, **25**, 1115-17 (1953).
- (119) Lopez, R. C., *Anales real acad. farm.*, **16**, 459-500 (1950).
- (120) Loscalzo, A. G., and Benedetti-Pichler, A. A., *Mikrochemie ver. Mikrochim. Acta*, **40**, 232-5 (1953).
- (121) Lourenço, O. B., *Anais assoc. quim. Brasil*, **10**, 57-61 (1951).
- (122) Ma, T. S., and McKinley, J. D., Jr., *Mikrochim. Acta*, **1953**, 4-13.
- (123) Mangeney, G., *Bull. soc. chim. France*, **1951**, 809-20.
- (124) Marsadro, M., *Mikrochemie ver. Mikrochim. Acta*, **38**, 372-5 (1951).
- (125) Mitsui, T., *Bull. Research Inst. Food Sci., Kyoto Univ.*, No. 8, 36-44 (1952).
- (126) Mitsui, T., *Japan Analyst*, **1**, 130-3 (1952).
- (127) *Ibid.*, **2**, 3-5 (1953).
- (128) Mitsui, T., and Tanaka, J., *J. Agr. Chem. Soc. Japan*, **24**, 188-90 (1950).
- (129) Nakamura, G. R., *ANAL. CHEM.*, **24**, 1372 (1952).
- (130) Nash, L. K., *Ibid.*, **23**, 1868-70 (1951).
- (131) Nogrády, G., *Magyar Kém. Lapja*, **4**, 350-2 (1949).
- (132) Nutten, A. J., *Mikrochemie ver. Mikrochim. Acta*, **39**, 355-9 (1952).
- (133) Oda, N., Kubo, M., and Norimasa, K., *J. Pharm. Soc. Japan*, **72**, 1079-81 (1952).
- (134) Ogg, C. L., *J. Assoc. Offic. Agr. Chemists*, **35**, 305-17 (1952).
- (135) *Ibid.*, **36**, 344-54 (1953).
- (136) *Ibid.*, pp. 335-44.
- (137) Ogg, C. L., and Willits, C. O., *Ibid.*, **35**, 288-91 (1952).
- (138) Otting, W., *Mikrochemie ver. Mikrochim. Acta*, **38**, 551-3 (1951).
- (139) Pagel, H. A., and Oita, I. J., *ANAL. CHEM.*, **24**, 756-8 (1952).
- (140) Parkin, B. A., Fernandez, J. B., Braun, J. C., and Rietz, E. G., *Ibid.*, **25**, 841 (1953).
- (141) Parks, T. D., *Anal. Chim. Acta*, **6**, 553-66 (1952).
- (142) Pepkowitz, L. P., *ANAL. CHEM.*, **24**, 900-1 (1952).
- (143) Peters, E. D., Rounds, G. C., and Agazzi, E. J., *Ibid.*, **24**, 710-14 (1952).
- (144) Peters, J. H., and Gutmann, H. R., *Ibid.*, **25**, 987-8 (1953).
- (145) Pirt, S. J., and Chain, E. B., *Biochem. J.*, **50**, 716-19 (1952).
- (146) Rao, D. S., and Shah, G. D., *Mikrochemie ver. Mikrochim. Acta*, **40**, 254-7 (1953).
- (147) Ribas, I., and Vázquez-Gesto, D., *Inform. quim. anal. (Madrid)*, **7**, 29-42 (1953).
- (148) Rogers, L. B., *J. Chem. Educ.*, **29**, 612-15 (1952).

- (149) Sahashi, Y., and Shibasaki, H., *J. Agr. Chem. Soc. Japan*, **25**, 57-9 (1951-52).
- (150) Sakamoto, S., *J. Pharm. Soc. Japan*, **72**, 509-10 (1952).
- (151) Salt, H. B., *Analyst*, **78**, 4-14 (1953).
- (152) Sarakhov, A. I., *Doklady Akad. Nauk S.S.S.R.*, **86**, 989-92 (1952).
- (153) Savacool, R. V., and Ullyot, G. E., *ANAL. CHEM.*, **24**, 714-17 (1952).
- (154) Scalamandre, A. A., and Guerrero, A. H., *Anales assoc. quim. Argentina*, **39**, 28-30 (1951).
- (155) Schaffer, F. L., Fong, J., and Kirk, P. L., *ANAL. CHEM.*, **25**, 343-6 (1953).
- (156) Schöniger, W., *Mikrochemie ver. Mikrochim. Acta*, **38**, 132-5 (1951).
- (157) *Ibid.*, **40**, 27-9 (1952).
- (158) Schöniger, W., *Mitt. Lebensm. Hyg.*, **43**, 105-16 (1952).
- (159) Schöniger, W., and Lieb, H., *Ibid.*, **38**, 165-7 (1951).
- (160) Schöniger, W., Lieb, H., and Gassner, K., *Z. anal. Chem.*, **134**, 188-91 (1951).
- (161) Schreiner, H., *Mikrochemie ver. Mikrochim. Acta*, **38**, 273-7 (1951).
- (162) Seligson, D., and Seligson, H., *J. Lab. Clin. Med.*, **38**, 324-30 (1951).
- (163) Sheers, E. H., and Cole, M. S., *ANAL. CHEM.*, **25**, 535 (1953).
- (164) Shelberg, E. F., *Ibid.*, **23**, 1492-3 (1951).
- (165) Shishikura, Y., *J. Japan. Biochem. Soc.*, **19**, 145-9 (1947).
- (166) Sickels, J. P., and Schultz, H. P., *Quart. J. Florida Acad. Sci.*, **14**, 35-7 (1951).
- (167) Singer, E., *J. phys. radium*, **12**, 554-62 (1951).
- (168) Sirotenko, A. A., *Mikrochemie ver. Mikrochim. Acta*, **40**, 30-1 (1952).
- (169) Slavík, K., and Smetana, R., *Chem. Listy*, **46**, 648-9 (1952).
- (170) Smith, D. M., Mitchell, J., Jr., and Billmeyer, A. M., *ANAL. CHEM.*, **24**, 1847-9 (1952).
- (171) Staemmler, H. J., *Biochem. Z.*, **323**, 74-7 (1952).
- (172) Steyermark, A., and Faulkner, M. B., *J. Assoc. Offic. Agr. Chemists*, **35**, 291-304 (1952).
- (173) Steyermark, A., and Garner, M. W., *Ibid.*, **36**, 319-35 (1953).
- (174) Steyermark, A., McNally, M. J., Wiseman, W. A., Niven, R., and Biava, F. P., *ANAL. CHEM.*, **24**, 589-90 (1952).
- (175) Stock, J. T., and Fill, M. A., *Mikrochim. Acta*, **1953**, 89-91.
- (176) Sugiyama, N., and Furuhashi, K., *J. Chem. Soc. Japan, Pure Chem. Sect.*, **72**, 583-5 (1951).
- (177) Sundberg, O. E., and Royer, G. L., *ANAL. CHEM.*, **24**, 907-8 (1952).
- (178) Szekeres, L., Fóti, G., and Pályi, E., *Magyar Kém. Folyóirat*, **56**, 377-8 (1950).
- (179) Thompson, R. C., *ANAL. CHEM.*, **25**, 535-6 (1953).
- (180) Thorn, J. A., and Shu, P., *Can. J. Chem.*, **29**, 558-62 (1951).
- (181) Throckmorton, W. H., and Hutton, G. H., *ANAL. CHEM.*, **24**, 2003-4 (1952).
- (182) Toennies, G., and Bakay, V., *Ibid.*, **25**, 160-5 (1953).
- (183) Unterzaucher, J., *Analyst*, **77**, 584-95 (1952).
- (184) Upson, U. L., *ANAL. CHEM.*, **25**, 977-9 (1953).
- (185) Vandenheuvel, F. A., *Ibid.*, **24**, 847-51 (1952).
- (186) Van Etten, C. H., *Ibid.*, **23**, 1697-8 (1951).
- (187) Van Etten, C. H., and Wiele, M. B., *Ibid.*, **25**, 1109-11 (1953).
- (188) White, L. M., and Morris, R. T., *Ibid.*, **24**, 1063-4 (1952).
- (189) Willits, C. O., *Ibid.*, **23**, 1565-7 (1951).
- (190) Wilson, C. L., *Mikrochim. Acta*, **1953**, 58-70.
- (191) Witekowa, S., *Wiadomości Chem.*, **4**, 174-89 (1950).
- (192) Wurzschildt, B., *Mitt. Lebensm. Hyg.*, **43**, 126-8 (1952).
- (193) Zacherl, M. K., and Stöckl, W., *Mikrochemie ver. Mikrochim. Acta*, **38**, 278-87 (1951).
- (194) Zimmermann, W., *Ibid.*, **40**, 162-9 (1952).